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Presentation and role of transplantation in adult patients with type 1 primary hyperoxaluria and the I244T *AGXT* mutation: Single-center experience

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Primary hyperoxaluria type 1 (PH1) is a rare genetic disorder characterized by allelic and clinical heterogeneity. We aim to describe the presentation and full single-center experience of the management of PH1 patients bearing the mutation described in our community (I244T mutation + polymorphism P11L). Since 1983, 12 patients with recurrent renal lithiasis have been diagnosed with PH1 and renal failure in the Canary Islands, Spain. Diagnostic confirmation was based on the presence of oxalosis in undecalcified bone or kidney allograft biopsy, reduced alanine:glyoxylate aminotransferase activity in liver biopsy, and blood DNA analysis. Patients underwent different treatment modalities depending on individual clinical circumstances and therapeutic possibilities at the time of diagnosis: hemodialysis, isolated kidney, simultaneous liver–kidney, or pre-emptive liver transplantation. In all cases, the presentation of advanced renal disease was relatively late (>13 years) and no cases were reported during lactancy or childhood. The eight patients treated with hemodialysis or isolated kidney transplantation showed unfavorable evolution leading to death over a variable period of time. In contrast, the four patients undergoing liver transplantation (three liver + kidney and one pre-emptive liver alone) showed favorable long-term allograft and patient survival (up to 12 years follow-up). In conclusion, in this PH1 population, all bearing the I244T mutation, the development of end-stage renal disease was distinctive during late adolescence or adulthood. Our long-term results support pre-emptive liver transplantation at early stages of renal failure, and kidney–liver transplantation for those with advanced renal disease.

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Primary hyperoxaluria type 1 (PH1) is a rare autosomal-recessive disorder characterized by a functional defect of the liver peroxisomal enzyme alanine:glyoxylate aminotransferase, which shows considerable phenotypic and genotypic heterogeneity.[1,2] Its estimated prevalence is over a broad range of 1–2.9 per million population.[3,4] In the Canary Islands, an Atlantic overseas region of Spain, we have observed a disproportionately high prevalence of PH1.[5,6] Strikingly, a large percentage of these patients are originally from one small island, that of La Gomera with approximately 17 000 inhabitants[5] (Figure 1). Recently, we have identified the Ile-244 → Thr (I244T) mutation in the *AGXT* gene in our PH1 patients or their relatives. In these cases, hyperoxaluria results from I244T mutation in combination with the common polymorphism Pro-11 → Leu (P11L).[7] This disorder constitutes an example of protein conformational disease, where misfolded proteins form functionally inactive aggregates.[7]

As this enzyme is only expressed in the liver,[2] this metabolic defect can only be treated by liver transplantation. In PH1 patients with end-stage renal disease (ESRD), simultaneous liver–kidney transplantation, first reported in 1987,[8] is currently considered the treatment of choice.[9–12] For patients without advanced renal disease, pre-emptive liver transplantation is the definitive treatment, although current experience is very limited and mainly reported in young children and infants.[13–17]

Our aim was to describe the full single-center experience in the management of adult patients with PH1 and renal disease during the last two decades. Of particular consideration, all our patients were late adolescents or adults, bearing the same genetic mutation, and were managed according to the therapeutic resources available at the time of diagnosis.

RESULTS

Clinical and genetic data, as well as patient evolution during dialysis or after transplantation, are shown in Tables 1 and 2. Family antecedents of renal stone formation were established for nine patients, and was doubtful or unknown in other patients. Parental consanguinity was verified in the three

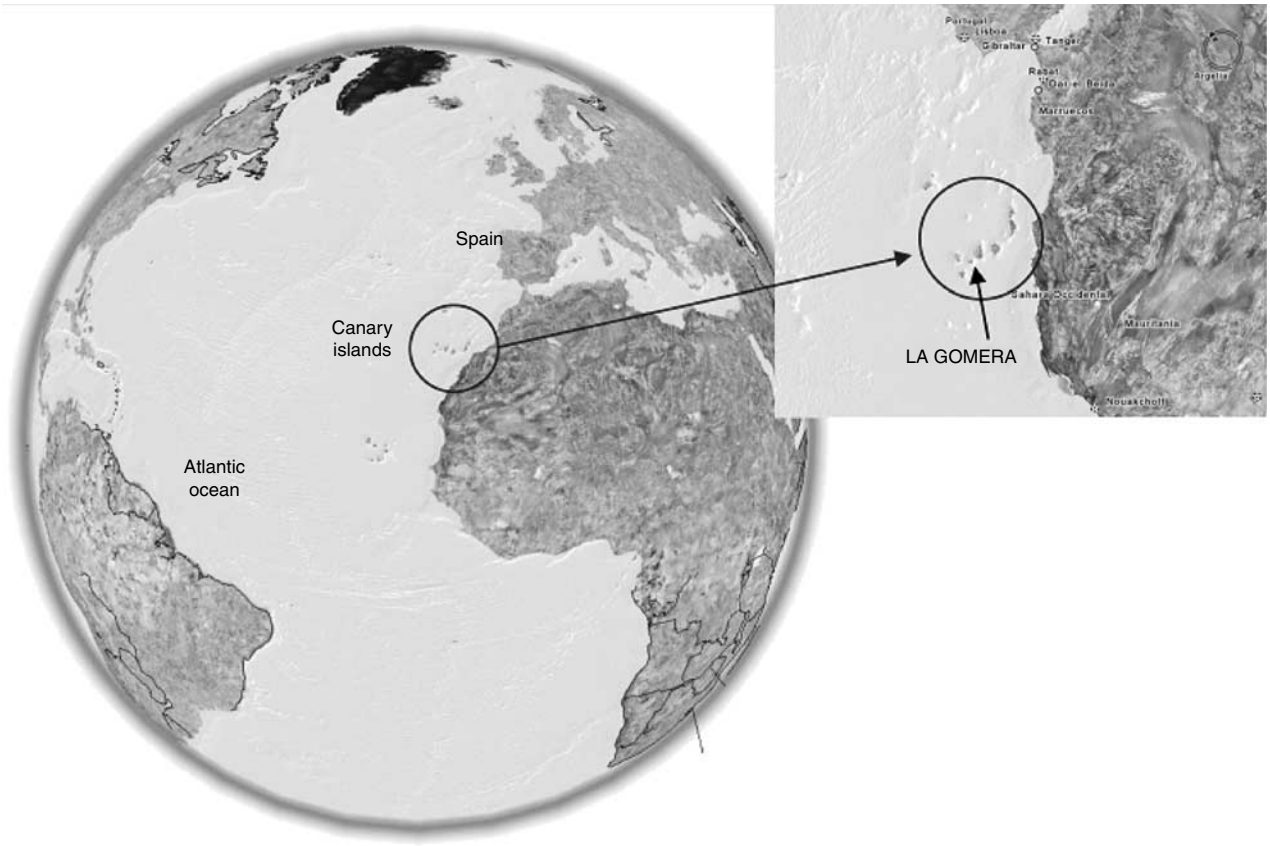

Figure 1 | Map showing geographic location of Canary Islands.

Table 1 | PH1 patients who did not receive liver transplant

Case	Age[a] Sex	Diagnostic confirmation	Mutation	Evolution	Cause of death
1	19 M	Bone biopsy	Homozygous I244T	Died after 1.5 years on HD	Oxalosis Complications
2	41 M	Bone biopsy	Unknown	Died after 8 years on HD	Oxalosis Complications
3	38 M	Bone biopsy	Homozygous I244T	KTx failed (relapse of oxalosis) Died after 7 years on HD	Oxalosis Complications
4	18 F	Bone biopsy KTx biopsy	Homozygous I244T	3 years on HD Died 8 months after KTx	Oxalosis+sepsis CMV
5	42 F	KTx biopsy[b]	Homozygous I244T	1 year on HD Died 3 months after KTx	Sepsis Pn Carinii
6	20 M	Bone biopsy	Homozygous I244T	2.5 years on HD Died 14 months after KTx	Oxalosis Complications
7	34 M	1st KTx biopsy[b]	Homozygous I244T	1 year on HD 1° KTx survived 5 years, relapse of oxalosis 2° KTx survived 8 years, relapse of oxalosis Died after 2 years on HD	Late oxalosis complications
8	60 F	KTx biopsy[b]	Compound heterozygous G170R and I244T	1 year on HD KTx failed, relapse of oxalosis	Still living on HD

CMV, cytomegalovirus; HD, hemodialysis; F, female; KTx, kidney allograft transplantation; M, male; PH1, primary hyperoxaluria type 1.
[a]Age at the initiation of HD in years.
[b]Diagnosed after KTx.

Table 2 | PH1 patients who received simultaneous liver–kidney (L–K Tx) or pre-emptive liver transplantation (LTx)

Case	Age[a] Sex	Diagnostic confirmation	Mutation	Vintage Date and type of Tx	Graft evolution (immunosuppression)
9	25 M	Brother of Case 1 Liver biopsy.	Homozygous I244T	2 years on HD 1993 L–K Tx	Functioning at 12 years Tacrolimus+prednisone sCre: 4.1 mg/dl
10	23 M	Brother of Case 1 Hyperoxaluria	Homozygous I244T	1994 Pre-emptive LTx	Functioning at 11 years Ciclosporine (monotherapy) sCre: 1.2 mg/dl
11	19 F	Liver biopsy	Homozygous I244T	1 year on HD 1997 L–K Tx	Functioning at 8 years Cyclosporine+prednisone sCre: 1.2 mg/dl
12	16 M	KTx biopsy[b]	Homozygous I244T	3 months on HD RTx failed. 18 months on HD 2001 L–K Tx	Functioning at 4 years Tacrolimus+prednisone+mycophenolate sCre: 1.1 mg/dl

HD, hemodialysis; F, female; KTx, kidney allograft transplantation; M, male; PH1, primary hyperoxaluria type 1; sCre, serum creatinine.
[a]Age at the time of transplantation (years).
[b]Diagnosed after KTx.

affected brothers (Patients 1, 9, and 10) and in two other cases (Patients 6 and 11).

The first clinical manifestations were renal colic and calculi passage in all patients except Patient 12, who commenced with nephrocalcinosis and renal failure. The age of onset was variable: lactancy and childhood in seven patients, adolescence and early adulthood in two cases, and during the fourth decade of life in the two remaining patients. Nephrocalcinosis was detected in all cases. Manifestations of obstructive uropathy occurred in most patients and seven patients required some kind of intervention, including surgery or lithotripsy. Hence, one patient underwent unilateral nephrectomy 1 year after liver–kidney transplantation (Patient 9) and another bilateral nephrectomy while on hemodialysis (Patient 6).

Hyperoxaluria was documented in four patients with variable degrees of renal failure (Patients 6 and 9–11). Patient 10 showed absolute resistance to pyridoxine and antilithogenic therapy, despite being treated at an early stage. In Patient 12, high urinary excretion of oxalate and glycolate was evident after the first kidney transplantation, which failed shortly thereafter.

Advanced renal failure developed relatively late in this population. There were no cases of ESRD during lactancy or childhood; one patient (number 12) initiated dialysis at 13 years of age and the rest during adulthood, from 18 up to 62 years of age.

Table 1 shows the outcome of eight patients treated with hemodialysis or renal transplantation alone. All six isolated renal transplants failed owing to ongoing oxalate production and recurrent disease. Most of these eight patients developed progressive bone disease, mimicking renal osteodystrophy but with inappropriate osteosclerosis and progressive disability. Clinical manifestation of systemic oxalosis appeared over a variable period of time. The main complications were bone and joint pain, polyneuropathy, and arrythmia presumed to be secondary to myocardial oxalate deposition. Systemic oxalosis was extremely severe in Patient 1, who evolved with bone deformities, opiaceous-dependent bone and joint pain, and absolute disability. Over the first 2 years on hemodialysis, the patient suffered ischemic necrosis of several phalanges, spontaneous loss of teeth, and diffuse extraosseous calcifications. Patient 12 showed retarded growth, responding minimally to growth hormone therapy before liver–kidney transplantation. As reflected in the table, the diagnosis of PH1 was confirmed by bone biopsy ($n = 6$) or renal allograft biopsy ($n = 2$).

Table 2 shows data of patients who received simultaneous liver–kidney ($n = 3$) or pre-emptive liver transplantation ($n = 1$). Patients who underwent liver–kidney transplantation received aggressive pre-transplant hemodialysis and there were no cases of post-transplant acute tubular necrosis. There was no long-term morbidity associated with the transplantation procedure. At the close of this study, all kidney and liver allografts were still functioning. Oxaluria remained below 40 mg/24 h in all cases and serum creatinine remained very close to the normal range in all patients, except Patient 9. This patient had urological complications – unrelated to PH1 – and chronic renal failure, but he remains free of dialysis after 12 years of follow-up.

DISCUSSION

PH1 is a rare genetic disorder characterized by allelic and clinical heterogeneity. For this reason, it is extremely difficult to establish a correlation between the genetic profile and the clinical presentation. Previous reports, mainly based on renal patient registries, have reported that PH1 presents in approximately 10% of affected individuals very early during lactancy or childhood, 80–90% in late childhood or early adolescence, and fewer than 10% of affected individuals present in adulthood.[17]

To date, over 50 different mutations and seven polymorphisms have been identified, and some of them have been related with either pyridoxine responsiveness or very early development of ESRD.[18–20] These studies have shown that mutation analysis may help to predict the clinical course and to define the best therapeutic strategy. In our community, we were able to follow patients with the same genetic mutation, and interestingly, recurrent renal lithiasis and ESRD appeared relatively late in this series, between the second and sixth decade of life. To our knowledge, cases of PH1 developing to advanced renal failure during lactancy or childhood have not been observed or reported by the relatives in our region. Our data suggest that this genetic variant of PH1 seems to be associated with severe renal stone disease and ESRD, but of relatively late presentation. However, we were unable to ascertain pyridoxine response in this mutation, as only one patient was attended at an early stage of renal failure (Case 10). This case showed an absolute resistance to doses of pyridoxine up to 15 mg/kg/day.

Strategies for treating patients with PH1 include reduction of oxalate production, antilithogenic measures, treatment of systemic oxalosis by dialysis and/or kidney transplantation, and cure of the metabolic defect by liver transplantation.

Management protocols in PH1 applying classic conservative measures have proved to be relatively effective in patients who have not yet developed advanced renal failure.[21–23] However, in patients with severe lithiasis and early renal failure, these measures are often insufficient and patients inevitably require renal replacement therapy. All authors agree that treatment with hemodialysis[6,23,24] is discouraging, particularly in severe cases. After isolated renal transplantation, recurrence of oxalosis is the norm,[6,24,25] although acceptable results have been reported in selected patients applying intensive protocols during the peri-transplant period.[22,26] These strategies should probably be reserved for less severe cases where irreversible renal failure appears after the fifth or sixth decades of life.

Liver transplantation is currently the treatment of choice to correct the underlying disease. To date, most liver transplantation has been performed after or simultaneously with renal transplantation.[9–12] The first pre-emptive liver transplant was performed in 1989.[13] Since then, several isolated cases have been reported with successful results, most of them in young children and infants.[13–17] To date, all available information on kidney–liver or pre-emptive liver transplantation is based on patient registries or case series.[17] Although published results are really encouraging, caution should be exercised because some PH1 patients receiving transplantation may be unreported, in particular those with unfavorable outcomes. The recent availability of an international registry for primary hyperoxaluria (http://mayoresearch.mayo.edu/mayo/research/nephrology/registry.cfm) should greatly improve data collection and analysis.

This single-center experience shows the complete outcomes (favorable and unfavorable) of different therapeutic modalities, applied to adult patients with advanced PH1 over different periods of time. Although rigorous comparison of therapeutic strategies is not possible, the frustrating results obtained using more conservative strategies clearly contrast with the favorable long-term outcomes observed in patients who underwent liver transplantation. One limitation of our study is that patients were diagnosed late in the course of the disease, when referred to our hospital. We can only speculate that early diagnosis and the application of prompt conservative measures might have affected the phenotype. Indeed, the cases with the favorable outcome were also those with the better diagnostic evaluation and more adequate treatment. Our results underscore that early diagnosis is crucial in PH1 patients to ensure proper treatment and the best outcome possible.

The form of PH1 described in our community constitutes a type of protein conformational disease, in which the synergistic effect of a common polymorphism (P11L) and a founder mutation (I244T) results in a misfolded protein that tends to form inactive aggregates.[7] In principle, this entity may benefit from new early cell-based therapies with pharmacological chaperones or small molecules to minimize protein aggregation.[7] Meanwhile, our long-term results in adult patients provide additional and strong support for an early inclusion in programs of pre-emptive liver transplantation or alternatively kidney–liver transplantation for those with advanced or ESRD.

MATERIALS AND METHODS

Since 1983, 12 patients have been diagnosed with PH1 and advanced renal failure in our hospital. We reviewed the clinical records of all 12 patients, the median age at initiation of hemodialysis was 29 years, ranging from 13 to 62 years; eight patients were men and four women, all Caucasian. At the time of diagnosis of PH1, two patients had moderate and advanced renal failure, respectively; six patients were on maintenance hemodialysis; and four patients had undergone renal transplantation. Patients were treated with different modalities according to individual clinical circumstances and therapeutic possibilities. Treatment alternatives were hemodialysis and kidney transplantation during the late 1980s, and simultaneous kidney–liver or pre-emptive liver transplantation when possible during the 1990s.

Diagnosis of primary hyperoxaluria

Given that most patients were studied and treated in the late 1980s and 1990s, diagnosis was established by the resources available at that time. Family history of renal colic, radiologic evidence of calcium nephrolithiasis and/or nephrocalcinosis, and radiologic osteosclerosis were the main diagnostic clues. Oxaluria detection was used in only one case with residual renal function.

The presence of diffuse oxalate deposits in undecalcified bone biopsy (Figure 2) or in kidney allografts, and decreased or absent alanine:glyoxylate aminotransferase activity demonstrated in the liver biopsy were the procedures used for diagnostic confirmation.

Undecalcified transiliac bone biopsies were obtained with Bordier trephine after double tetracycline labeling applying a standard technique.[27] At that time, using this technique to study renal osteodytrophy, we detected our first cases of PH1.

Alanine:glyoxylate aminotransferase enzymatic activity determined in liver biopsies[28] was performed at a reference center

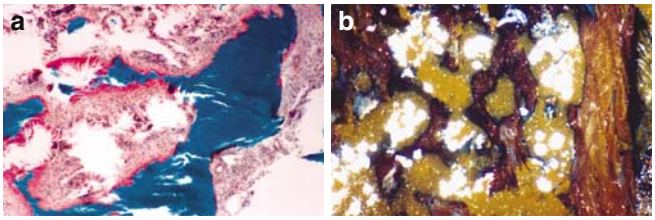

Figure 2 | Transiliac undecalcified bone specimen. (**a**) Marrow granulomatous reaction constituted by multinucleated giant cells, fibroblasts, and fibrosis (Masson–Goldner staining; out of focus areas are hard to avoid in non-decalcified bone sections). (**b**) Highly birefringent oxalate crystal deposition grouped in stars or rosette (Toluidine blue staining viewed under polarized light).

(Department Chemical Pathology, University College of London) for diagnostic confirmation (Cases 9 and 11).

Blood DNA was obtained from all patients and their relatives when possible. In patients who had died, relatives were contacted to obtain blood samples. DNA analysis was performed as described.[5] In one deceased patient (Case 2), genetic information was not available. However, the patient came from the same region, which suggests that he had the same genetic profile.

Informed consent to perform liver and/or kidney transplant was obtained after a complete explanation of therapeutic options, risks, and advantages.

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